BILIRUBIN TOTAL
Hitachi® Liquid Reagent

KIT SPECIFICATIONS:

Cat. No.	*Quantity*	*Reagent*	*Storage*
AD062	8 x 40 ml	TBIL 1	2-8°C
	4 x 20 ml	TBIL 2	

INTENDED USE:

In Vitro Diagnostic reagent pack for the quantitative determination of Total Bilirubin in serum and plasma on Hitachi® automated analysers.

SUMMARY AND EXPLANATION: [1]

Serum Bilirubin is present in 4 different isomers. Unconjugated α, monoconjugated β, diconjugated γ and covalently albumin bound δ. Bilirubin is produced from the degradation of red blood cells. It is extracted and biotransformed in the liver and excreted and the bile and urine. Bilirubin measurements are made primary for the diagnosis of liver diseases and the detection of haemolytic anaemia.

PRINCIPLE OF THE TEST: [2]

Indirect Bilirubin is liberated by the detergent. Total Bilirubin is, then, coupled with a diazonium compound to give the corresponding azobilirubin which is measured photometrically. The concentration of the compound formed is proportional to the total Bilirubin concentration in the sample.

Bilirubin + diazonium ion $\xrightarrow{pH<2}$ azobilirubin

WARNINGS AND PRECAUTIONS:

For In Vitro Diagnostics Use Only - For Professional Use Only
Carefully read instructions for use. Deviations from this procedure may alter performance of the assay.

Components Colour and Appearance:
Reagent 1: Clear, colourless liquid.
Reagent 2: Clear, pale beige liquid
Any significant changes from the above could indicate that the assay might be compromised. Refer to Laboratory's QC program for actions to be taken. In case of serious damage to the bottle and/or cap, resulting in product leakage and/or contamination, do not use the reagent pack and contact your distributor.

Safety precautions:
CAUTION: Take all necessary precautions required when handling laboratory reagents. Material Safety Data Sheet is available upon request.

Handling precautions:
- Do not use components past the expiry date stated on the Bottles.
- Do not Freeze Reagents.
- Do not use components for any purpose other than described in the "Intended Use" section.
- Do not interchange caps among components as contamination may occur and compromise test results.
- Refer to local legal requirements for safe waste disposal.
- **Reagent 2 is light sensitive and must be stored in the dark**

INSTRUMENTS:

This assay is designed to run on Hitachi® clinical chemistry analysers. Refer to relevant user's manual or Laboratory internal practice for routine maintenance procedures. *All information is encoded in the barcode. If analyser fails to read or if the barcode is damaged, enter the series of numbers beneath the barcode.*

COMPONENT COMPOSITION:

Component	*Ingredients*	*Concentration in Tests*
Reagent 1	Phosphate buffer	40 mmol/l
	(NaCl)	9 g/l
Reagent 2	2,4-Dichlorophenyl-diazonium salt	1 mmol/l
	HCl	30 mmol/l

REAGENT PREPARATION AND STABILITY:

Reagent 1 and 2 are ready for use.
Before use, mix reagent by gently inverting each bottle.
If stored and handled properly, unopened component is stable until expiry date stated on the label.
Stability On Board the Instrument: 28 days.
Reagent 2 is light sensitive and must be stored in the dark

TYPE OF SPECIMEN: [1]

Use serum, free of haemolysis, or EDTA / heparinised plasma as specimen.
A morning specimen from a fasting patient should be preferred.

It is recommended to follow NCCLS procedures (or similar standardised conditions) regarding specimen handling. Specimen should be collected in an appropriate sample container, with proper specimen identification. Plasma/Serum should be separated from cells within 2 hours after collection.
Stability: It is essential to store specimens in the dark, at 2-8°C. Only in those conditions, stability is maintained for 3 days.

TEST PROCEDURE:

Materials required but not supplied:

Description	*Catalog. No.*	*Description*	*Catalog. No.*
General Chemistry Calibrator	AD983	Hitachi® Analyser	N/A
General Chemistry Control Level 1	AD922	Hitachi® Consumables	N/A
General Chemistry Control Level 2	AD932	General Laboratory Equipment	N/A

Assay procedure:
Refer to relevant user's manual for instructions on instrument start-up, loading components and samples, calibration, sample testing procedures, calculating and reporting results.

Calibration:
Using recommended Calibrator, calibrate the assay:
- When using a new reagent kit or changing lot number.
- Following preventive maintenance or replacement of a critical part.
- When Quality Controls are out of range.

Quality Control:
All clinical laboratories should establish an Internal Quality Control program. Verify instrument and reagent performance with recommended controls or similar. The values obtained for QC should fall within manufacturer's acceptable ranges or should be established according to the Laboratory's QC program.
Controls should be assayed:
- Prior to reporting patient results.
- Following any maintenance procedure.
- At intervals established by the laboratory QC Programme.

CALCULATION:

The analyser automatically calculates the Total Bilirubin concentration in the sample.
(Conversion Factor: Qty in µmol/l = 17.1 x Qty in mg/dl.)

EXPECTED VALUES: [1]

	µmol/l	*mg/dl*
Serum	0 - 21	0-1.2

Each laboratory should establish its own reference range. Total Bilirubin results should always be reviewed with the patient's medical examination and history.

PERFORMANCE CHARACTERISTICS:

Performance results can vary with the instrument used. Data obtained in each individual laboratory may differ from these values.

Linearity:
This assay is linear up to 513 µmol/l (30mg/dl).
For samples with a higher concentration:
- Reassay using, when available, "Rerun" function. Refer relevant user's manual for instructions.
- *Or*, dilute 1:1 with 0.9% NaCl (9g/l) and re-assay. Multiply result obtained by 2.

Interfering substances:
Results of study are as follows:

Haemolysis: Less than 10% interference up to 2.5 g/l Haemoglobin
Lipemia: Less than 10% interference up to 5 g/l Intralipid.

Sensitivity:
The Lowest Detectable Level was estimated at 1.2 µmol/l.

Precision:

Intra-assay N = 20	Mean *(mg/dl)*	*SD*	*% CV*	*Inter-assay N = 20*	Mean *(mg/dl)*	*SD*	*% CV*
Level 1	1.02	0.02	2.32	Level 1	1.15	0.04	3.49
Level 2	4.83	0.05	0.95	Level 2	4.65	0.13	2.86

Method Comparison:
Using 50 samples, a comparison, between this Total Bilirubin DPD test (y) and another commercially available test (x), gave the following results:

y = 1.00 (x) - 0.00mg/dl	r = 1.000	Sample Range from 2 to 228 µmol/l

BIBLIOGRAPHY:

1. Burtis CA, Ashwood ER. Tietz Fund. Of Clin. Chem. 5th ed. 30-54, 601-606 and 966.
2. Weigh E, Bach H, Kreig US. Med. Klin. 1975; 70: 664.

SYMBOLS:

The following symbols are used in the labelling of Audit Diagnostics systems:

IVD	In Vitro Diagnostics	REF	Catalogue No
LOT	Batch Code	CONT	Content
REAG	Reagent	Ab	Antibody
CAL	Calibrator	SUBS	Substrate
BUF	Buffer	CE	CE Mark - Device complies with the Directives 98/79/EC
	Storage temperature	→	Reconstitute with
	Expiry Date (Last day of the month)		Manufactured By
	Biological risk		Consult Instruction for Use

Manufactured By: AUDIT DIAGNOSTICS, Business & Technology Park, Carrigtwohill, Co. Cork (Ireland)
Tel: 00353 - (0) 21 – 4533 652 *Fax:* 00353 - (0) 21 – 4533 653
E-mail: info@auditdiagnostics.ie *Website:* www.auditdiagnostics.ie

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BILIRUBIN TOTAL Hitachi ® Instruments Settings

Catalogue No(s): AD062

Audit Diagnostics

HITACHI 911 / 912 / 917 / MODP®

All information is encoded in the barcode. If analyser fails to read or if the barcode is damaged, enter the series of numbers beneath the barcode.

HITACHI 704®: (Temperature: 37°C)

CHEMISTRY PARAMETERS	
TEST CODE	**[BIL-T]**
ASSAY CODE	[2(2POINT)] - [15] - [32]
SAMPLE VOLUME	[6]
R1 VOLUME	[240] - [__] - [NO]
R2 VOLUME	[60] - [__] - [NO]
WAVELENGTH	[700] - [546]
CALIB. METHOD	[LINEAR]
STD (1) CONC. POS.	[__] - [__]
STD (2) CONC. POS.	[__] - [__]
STD (3) CONC. POS.	[0] - [0]
STD (4) CONC. POS.	[0] - [0]
STD (5) CONC. POS.	[0] - [0]
STD (6) CONC. POS.	[0] - [0]
UNITS	[__]
SD LIMIT	[0.1]
DUPLICATE LIMIT	[100]
SENSITIVITY LIMIT	[0]
ABS LIMIT (INC/DEC)	[0] - [0]
PROZONE LIMIT	[32000] - [1]
EXPECTED VALUE	[__] - [__]
INSTRUMENT FACTOR	[1.00]

[__] User Defined.

HITACHI 717®: (Temperature: 37°C)

CHEMISTRY PARAMETERS	
TEST CODE	**[BIL-T]**
ASSAY CODE	[2(2POINT)] - [24] - [50]
SAMPLE VOLUME	[6] - [__]
R1 VOLUME	[240] - [__] - [NO]
R2 VOLUME	[60] - [__] - [NO]
WAVELENGTH	[700] - [546]
CALIB. METHOD	[LINEAR]
STD (1) CONC. POS.	[__] - [__]
STD (2) CONC. POS.	[__] - [__]
STD (3) CONC. POS.	[0] - [0]
STD (4) CONC. POS.	[0] - [0]
STD (5) CONC. POS.	[0] - [0]
STD (6) CONC. POS.	[0] - [0]
SD LIMIT	[0.1]
DUPLICATE LIMIT	[500]
SENSITIVITY LIMIT	[0]
ABS LIMIT (INC/DEC)	[0] - [Inc]
PROZONE LIMIT	[0] - [Lower]
EXPECTED VALUE	[__] - [__]
PANIC VALUES	[__] - [__]
INSTRUMENT FACTOR	[1.00]

[__] User Defined.

HITACHI 911/912®: (Temperature: 37°C)

CHEMISTRY PARAMETERS		
TEST CODE		**[BIL-T]**
ASSAY CODE		[2 POINT END] - [10]
WAVELENGTH (SUB-MAIN)		[700] - [546]
ASSAY POINT		[15] - [31]
DILUTION		[3] - [14]
SAMPLE VOLUME (µL)		[6] - [__]
ABS LIMIT		[0] - [INCREASE]
PROZONE LIMIT		[0] - [Lower]
REAGENT (µL)	R1	[240] - [0] - [0]
	R2	[0] - [0] - [0]
	R3	[60] - [0] - [0]
	R4	[0] - [0] - [0]
CALIBRATION TYPE		[LINEAR] - [2] - [2]
SD LIMIT		[0.1]
DUPLICATE LIMIT		[500]
SENSITIVITY LIMIT		[240]
SI ABS. LIMIT		[-32000] - [32000]
UNIT		[__] - [__]
INSTRUMENT A		[1.00]
FACTOR (Y=AX+B) B		[0] - [0]
STD 1		[__] - [__]
STD 2		[__] - [__]
STD 3		[0] - [0]
STD 4		[0] - [0]
STD 5		[0] - [0]
STD 6		[0] - [0]

[__] User Defined.

HITACHI 902®: (Temperature: 37°C)

CHEMISTRY PARAMETERS		
TEST NAME		**[TBIL]**
ASSAY CODE		[2POINT END] - [10] - [__]
ASSAY POINTS		[17] - [21] - [0] - [0]
WAVELENGTH (SUB-MAIN)		[700] - [546]
SAMPLE VOLUME (µL)		[6]
REAGENT (VOL-POS-BOTTLE SIZE)	R1	[240] - [__] - [__]
	R2	[0] - [__] - [__]
	R3	[60] - [__] - [__]
CALIBRATION		[LINEAR] - [2] - [2]
CALIB 1 (CONC/POS)		[__] - [__]
CALIB 2 (CONC/POS)		[__] - [__]
CALIB 3 (CONC/POS)		[0] - [0]
CALIB 4 (CONC/POS)		[0] - [0]
CALIB 5 (CONC/POS)		[0] - [0]
CALIB 6 (CONC/POS)		[0] - [0]
S1 ABS		[0]
K FACTOR		[12000]
K 2 FACTOR		[10000]
K 3 FACTOR		[10000]
K 4 FACTOR		[10000]
K 5 FACTOR		[10000]
A FACTOR		[0]
B FACTOR		[0]
C FACTOR		[0]
SD LIMIT		[0.1]
DUPLICATE LIMIT		[500]
SENSITIVITY LIMIT		[__]
S1 ABS. LIMIT (LOW/HIGH)		[-32000] - [32000]
ABS LIMIT		[0] - [INCREASE]
PROZONE LIMIT		[0] - [Lower]
PROZONE (END POINT)		[35]
EXPECTED VALUES		[__] - [__]
INST FACTOR (A - B)		[1] - [0]
KEY SETTING		.....

[__] User Defined.

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